

POLICY ON RODENT SURGERY AND POST-OPERATIVE CARE

PURPOSE:

The purpose of this document is to outline procedures for aseptic surgery and post-operative care for rodents. The U.S. Public Health Service *Guide for the Care and Use of Laboratory Animals* states that survival surgery on rodents should be performed in facilities intended for that purpose, using sterile instruments, surgical gloves, and aseptic procedures to prevent clinical infections. Aseptic technique is used to reduce microbial contamination to the lowest possible practical level. Judicious use of anesthetic and analgesic agents to minimize any pain associated with the experimental procedure is scientifically and ethically imperative. This document should allow all personnel involved to understand what constitutes aseptic technique and what is considered to be appropriate pre- and post-operative care.

1) Non-Survival Surgery

- a) All animals must be sufficiently anesthetized prior to surgery such that they are completely unconscious and show no reaction to pain-eliciting procedures. For most rodents, a lack of response to toe pinching is indicative of surgical anesthesia. Refer to the Center for Comparative Medicine guidelines for anesthesia for appropriate anesthetic agents and dosages (<http://www.healthsystem.virginia.edu/internet/ccm/>).
- b) All animals are euthanized before recovery from anesthesia.
- c) All personnel handling the animals must wear gloves.
- d) Instruments and work surfaces need not be sterile but must be clean.
- e) The surgical site should be free of hair.
- f) Expired anesthetic and analgesic drugs **may not** be used for non-survival procedures. Other expired drugs can be used but should be specifically labeled “Non-survival Procedures Only” for this use.

2) Survival Surgery - Aseptic Technique

- a) Surgery must be conducted on a clean, uncluttered lab bench or table surface. The surface should be covered with a clean drape.
- b) Hair must be removed from the surgical site using clippers, hair plucking, or a depilatory agent. The surgical site must at a minimum be treated with a 70% ethyl alcohol wipe to cleanse the skin followed by an antiseptic solution (e.g. povidone iodine or chlorhexidine solution). A complete (and ideal) sterile prep is 3 alternating wipes with a disinfectant scrub and alcohol (or a sole agent such as Hibiclens), followed by application of antiseptic solution. Placing a sterile drape over portions of the body that are not aseptically prepared is recommended but not required.
- c) All instruments must be sterilized, with the method of choice based on the physical characteristics of the material to be sterilized. Sterile Supply located in the new hospital will autoclave or gas sterilize instruments that are properly wrapped. The Center for Comparative Medicine can also steam sterilize upon request. Sterilization indicators (heat sensitive tape) should always be placed inside the wrapped pack to confirm that the materials reached the appropriate temperature. Write the sterilization date on the outside

and use within 6 months or resterilize. Hot bead sterilizers are inexpensive and very convenient for bench top use, especially for performing surgery on multiple animals consecutively. Acceptable techniques for cold sterilization include soaking in 2% glutaraldehyde for 10 hours or 8% formaldehyde + 70% ethyl alcohol for 18 hours. There are several commercially available germicidal agents that are safer to use, but attention must be paid to the shelf life of solutions once they are prepared. Gauze pads, intravascular catheters, suture material, etc. should also be sterile. Please note placing instruments in 70% alcohol alone is **not** acceptable. Supplies obtained from "Surplus" that have been previously opened should be re-sterilized.

- d) The surgeon must wash his/her hands with an antiseptic surgical scrub preparation and then aseptically put on gloves. If working alone, the surgeon should have the animal anesthetized and positioned and have at least the first layer of the wrapped instruments or cold pack opened before putting on sterile gloves, so that he/she can remain sterile. Opened sterile items should be set down on a sterile surface, not just a blue pad or bench paper.
- e) The surgeon must wear a head cover and facemask. A sterile gown is recommended, but a clean scrub top, lab coat or nonsterile gown worn over street clothes is acceptable.
- f) Ophthalmic ointment must be administered in both eyes of the anesthetized rodent to protect the corneas from drying and abrasion.

3) **Multiple Animal Surgeries**

If performing surgeries on more than one animal consecutively, START WITH A STERILE INSTRUMENT PACK. The instruments can be placed in 70% ethyl alcohol in-between animals or preferably re-sterilized in a glass bead sterilizer. The alcohol should be replaced when moderately contaminated with blood or other body fluids. Sterile gloves should be changed between surgeries if the surgeon touches non-sterile surfaces. The surgeon may wipe his/her gloves for 30 seconds with sterile gauze pads soaked in alcohol between animals that are already prepped by another individual.

4) **Wound Closure**

- a) The abdominal or thoracic body wall should be closed with absorbable suture material. The skin should be closed with staples, wound clips, surgical adhesive, or with a non-absorbable suture material in a simple interrupted pattern. Do not use silk for skin closure. A subcuticular closure can be used in larger rodents. Non-absorbable skin sutures or staples should be removed when adequate healing is apparent, typically 7 to 10 days after surgery. If the survival time post-surgery is ≤ 14 days, suture removal is not necessary. The wound should be observed for swelling, heat, discharge and opening of the incision (dehiscence) at least once daily and treated with an antimicrobial agent if an infection is noted. NOTE: The suture acquired from Hospital Surplus may have expired and not be suitable for use for survival procedures.

5) **Post-operative Management**

- a) Rodents should be kept warm with an external heat source both during surgery and afterwards until ambulatory. A heating pad underneath the drape or a heat lamp can be used. Be careful not to overheat the rodent when using a lamp. Monitoring rectal body temperature is the best method to ensure that the animal is neither hypo- nor

hyperthermic. Rodents under anesthesia become essentially ectothermic and unable to control their body temperature. Hypothermia significantly prolongs anesthesia and impairs recovery.

- b) An analgesic agent must be administered prior to recovery from anesthesia. Preemptive pain management is much more effective than giving medication after observing signs of pain. **Consult with a veterinarian when writing the protocol to devise the most appropriate pain management plan for your experiments.** In general, researchers have a choice of 3 drug classes: an opiate such as buprenorphine, butorphanol, or morphine, a nonsteroidal anti-inflammatory (NSAID) such as ketoprofen, ketorolac, or carprofen, and/or a long-acting local anesthetic such as 0.25% bupivacaine. Buprenorphine at 0.1 mg/kg IM or SC works well for many surgical procedures because it provides analgesic blood levels for a period of 8-12 hours. Ketoprofen at 2 mg/kg SC once every 24 hours is a good first choice NSAID. If systemic analgesia is not suitable for scientific reasons or if the surgical intervention is minimal and expected to produce only mild short-lasting pain, local anesthesia such as bupivacaine (Marcaine) infiltration at the surgical incision may be an option. Oral dosing in the drinking water is generally not an effective method. Depending on the severity of trauma associated with the procedure, administration of an analgesic agent for up to 72 hours may be indicated.
- c) Antibiotics are not necessary if sterile technique is used. However, some procedures warrant the use of peri-operative antibiotics. This can be discussed with a veterinarian.
- d) After surgery, once the animal can freely move around the cage, it may be returned to the vivarium (return to barrier facilities is not permitted. Check with the facility supervisor for appropriate locations).
- e) At least once daily the animals should be observed for signs of pain, distress, and incision problems. The following list (from *Recognition and Alleviation of Pain and Distress in Laboratory Animals*, NRC 1992) provides various signs to be watched for that would indicate acute pain in rodents.
- i. Decreased appetite. May eat bedding or their offspring.
 - ii. Decreased urine and fecal output.
 - iii. Decreased activity.
 - iv. Piloerection, ungroomed appearance.
 - v. Excessive licking and scratching that may progress to self-mutilation.
 - vi. Abnormal stance or hunched posture.
 - vii. Respiration can be rapid and shallow with grunting or chattering on expiration.
 - viii. Pupils might be dilated.
 - ix. Porphyrin secretion (“red tears”) might be seen around the eyes and nose.
 - x. Vocalization.
 - xi. Increased aggressiveness when handled.
- f) Records must be kept of all surgeries performed, anesthesia and analgesia administered, and any complications encountered. If any of the above signs of pain or distress are observed, a staff member in Center for Comparative Medicine should be notified immediately so that veterinary care is provided. If the pain or distress is significant and intractable, the animal will be euthanized. See also the ”Policy and Guidelines on Humane Endpoints” and “Policy on Recognition and Assessment of Pain and Distress”. <http://www.healthsystem.virginia.edu/internet/iacuc/policies/policies.cfm>